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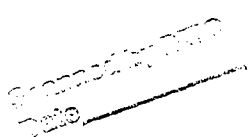
WADC TECHNICAL REPORT 53-58

ACOUSTIC TRAUMA IN THE GUINEA PIG

HALLOWELL DAVIS
CENTRAL INSTITUTE FOR THE DEAF

MARCH 1953



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Hallowell Davis
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FOREWORD

This report was prepared by Dr. Hallowell Davis and his collaborators at Central Institute for the Deaf, St. Louis, Missouri under United States Air Force Contract No. AF 18-(600)-58 with Washington University, St. Louis, Missouri, RDO No. 695-63, "Action of Vibration (Sonic and Mechanical) on Air Force Personnel." This contract was administered under the direction of the Bio-Acoustics Unit, Biophysics Branch, Aero Medical Laboratory, Wright Air Development Center with Captain William J. Gannon as Project Engineer. The experiments were carried out under the administrative supervision of the Principal Investigator, Dr. Walter P. Covell.

Dr. Davis was assisted in the experimental work on which this report is based by R. W. Benson, W. P. Covell, C. Fernandez, R. Goldstein, Y. Katsuki, J. P. Legouix, D. R. McAuliffe and I. Tasaki.

ABSTRACT

The ears of anaesthetized guinea pigs were exposed to intense tones of 185, 545, 2000 or 8000 cps at the sound pressure (at the eardrum) from 138 to 148 db re 0.0002 microbar. The electrical output was recorded by differential electrodes before, during and after the exposures.

The injury caused by 8000 cps centers in the basal turn, by 2000 cps in the second turn, by 500 and 185 cps in the third and fourth turns of the cochlea. No single tone, at the intensities and durations employed, injured all of the cochlea.

The "threshold" of the cochlear microphonic and also its maximum voltage are valid indices of the anatomical injury seen under the microscope if the proper test frequencies and positions of electrodes are used. Electrical assessment of injury will expedite future studies of acoustic trauma.

The destructiveness of a tone does not depend on its frequency. Equally severe (probably permanent) injuries were produced by a one-minute exposure at all four frequencies tested when the intensity level was about 144 db at the eardrum.

PUBLICATION REVIEW

This report has been reviewed and is approved.

FOR THE COMMANDING GENERAL:

Robert H. Blount

ROBERT H. BLOUNT
Colonel, USAF (MC)
Chief, Aero Medical Laboratory
Research Division

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INTRODUCTION

The present study of the physiological and anatomical effects of very loud sounds on the ears of guinea pigs was undertaken in conjunction with studies already in progress (1) that depend on anatomical methods alone. One object was to determine the relation between structural injury and the electrical output of the cochlea, i.e. the cochlear microphonics and the action potentials of the nerve. Previous studies of this sort (2,3,4) have given rather unsatisfactory correlations, but we hoped that our recently developed methods of inserting electrodes into the different chambers of the cochlea (5,6,7,8) would give sufficiently localized electrical pick-up to clarify the relationships and thus allow the use of the quicker electrical methods for establishing limits of tolerance of the inner ear with respect to the intensity, frequency and duration of exposure.

A second objective of this study was to find how the intensity of sound necessary to produce serious injury to an ear in a short time depends on the frequency of the sound. A preliminary study (9) had indicated that the frequency of 9000 cps was very effective in reducing the electrical output of the ear.

SECTION I

APPARATUS, METHODS AND CALIBRATIONS

Young adult guinea pigs, 200 to 300 gm body weight, were anaesthetized with dial (Ciba) in urethane (0.50 cc per kilo, intraperitoneally). The bulla was opened by a ventro-lateral approach (5) and small holes were drilled into one or more turns of the cochlea. Fine enamel-insulated wires with tips scraped bare were inserted into these holes. Small droplets of lacquer ("Radio Service Cement") on the wires prevented too deep penetration and greatly reduced any escape of perilymph. In our early experiments silver wire (diameter about 100 microns) was used but smaller nichrome-steel wire (about 20 microns, including the enamel insulation) has proved much more satisfactory. There is far less operative injury from the smaller holes and no risk of the toxic effects of silver, and our cathode-follower preamplifier (8) eliminates the difficulties of high electrode impedance. In the later experiments two pairs of electrodes were regularly placed, one pair in scala vestibuli and scala tympani of Turn I (basal) and the other pair in either Turn II, III or IV. (For Turns III and IV the extreme apex at the helicotrema is a satisfactory substitute for scala tympani, which is anatomically inaccessible in these turns. Scala media was always carefully avoided.)

The opening in the bulla was not closed with dental cement (5) but only by drawing the muscles and skin together, in order to facilitate inspection of the electrodes and removal of any serum or perilymph that might accumulate in the bulla. Other details of our operative methods, of our three-channel electrical recording system and of our electro-acoustic equipment for the generation of pure tones or "tone-pips" (brief amplitude-modulated wave trains) have been published elsewhere (8,10). Tone-pips set off the action potentials in well-synchronized easily-identified volleys.

We could thus observe simultaneously 1) the cochlear microphonic (CM) from a segment of organ of Corti about 2 mm long in the middle of the basal turn (7,8), 2) the cochlear microphonic from a similar segment of one or another

of the upper turns, and 3) action potentials (AP) from all of the cochlea, recorded from the entire auditory nerve as it passes through the internal auditory meatus (11). Our balanced input circuits (8) for "cancellation" of either CM or AP allowed us to observe and measure either AP or CM without significant contamination, in a normal ear, by the other. The tone-pip as a stimulus was very useful after acoustic trauma in differentiating AP from CM by virtue of the known latency of AP relative to the first negative wave of CM. This latency was sometimes the only clear basis for recognizing as AP a small response to a 2000 cps tone-pip after the local CM had been abolished by acoustic trauma.

Three kinds of electrical measurement were usually made before and again after the exposure to high-intensity sound:

1) The "threshold" of the cochlear microphonic for several frequencies of tone and tone-pips. The "threshold" response is an arbitrary small output of about 2 microvolts (rms), which can just be distinguished reliably from the noise level of the electrical system.

2) The threshold for action potentials in response to tone-pips of several frequencies delivered at a pulsing frequency of about 10 per sec. This threshold is also an arbitrary just-visible response.

3) The maximum voltage of the cochlear microphonic at several frequencies and the intensities of the tones at which the maximum output was reached; or, if the maximum was not reached at a "safe" level, the voltage output at an arbitrary high intensity, about 125 db re 0.0002 db at the eardrum.

Each of these measures was taken for three to five different frequencies, and from them were reckoned in decibels the losses of sensitivity or of output, due to the exposure. The measurements were repeated after an hour's recovery and usually again after a second hour, and any trend toward recovery or toward further deterioration of response was noted.

In many experiments we waited for one or two hours after inserting electrodes and speculum before exposing the ear to intense sound. Repeated measurements of thresholds and of maximum electrical outputs during this period assured us that the drilling of holes into scala vestibuli and scala tympani and the placement of electrodes in them did not cause deterioration of the action potentials or the cochlear microphonics, even when there was an appreciable leakage of

perilymph. The only requirements for stability of thresholds seemed to be that the animal's respiration should continue normally, that for acoustic reasons no large quantity of serum or perilymph should be allowed to remain in the middle ear or bulla, and that injury to the scala media should be scrupulously avoided. The animal was warmed if necessary to prevent shivering. Many of the animals served for other experiments lasting as long as five hours, with stable thresholds, before the acoustic trauma was inflicted.

The wave forms of the electrical responses of the cochlea were photographed on the oscilloscope during the traumatic exposures. They showed dramatically the extreme distortions produced by mechanical overload of both the middle ear and the inner ear and by the non-linearity of electrical output, but we have not analyzed them in detail.

Acoustic trauma was produced by the same electro-acoustic system that was employed for measuring the physiological thresholds before and after exposure. An Atlas PM-25 loudspeaker was connected by a rubber garden hose to a tight-fitting ear-speculum that was sewed securely into the animal's external auditory canal. The voltage at the terminals of the loudspeaker was observed throughout each exposure. It rarely fluctuated by more than ± 0.3 db. Originally the system was acoustically calibrated with a 640-AA microphone and a 1 cc coupler. The frequency characteristic showed strong peaks of resonance and at some of the peaks we could obtain pressures above 150 db* without electrical or mechanical overload. The peaks at or very near 185, 545, 2000 and 8000 cps were selected as the standard frequencies for traumatic exposure. The frequency was adjusted to the exact frequency of the resonant peak for each exposure and also for each calibration. The peak near 2000 cps, for example, averaged 1990 cps with a range from 1975 to 2000 cps. As test frequencies the conventional multiples and submultiples of 1000 cps were usually employed.

Later we succeeded in making reliable measurements of the acoustic pressure developed just in front of the eardrum in the ear canal of the animal. A probe-tube microphone made from a 20-gauge spinal puncture needle, 10 cm long, was employed (12). The needle was introduced surgically through the side of the canal in such a way as not to disturb the usual placement of the speculum or create an acoustic leak.

*Throughout this paper the acoustical reference level is 0.0002 microbar.

Most of the calibrations were made on animals immediately after death to facilitate accurate maintenance of the position of the tip of the probe tube. The resonant peaks and valleys were much less pronounced than when they were measured with the rigid 1 cc coupler, and the pressure developed in the upper part of the frequency range was found to be much higher at the eardrum than in the coupler. Table 1 shows the relations between the voltage applied to the terminals of the loudspeaker, the pressures measured in the 1 cc coupler and the pressures in the ear canal at the eardrum. The relationships between the pressures at the four different frequencies are quite constant for the five animals studied. Their rank order is always the same and the highest (at 545 cps) is always between 6.1 and 8.3 db greater than the lowest (at 8000 cps). The average level from animal to animal varies significantly with a range of 6.6 db between the extremes. The range for the most variable frequency (7.7 db at 545 cps) is only slightly greater than for the average. We have not yet found the reason for the variation in average level from animal to animal. As long as it exists we cannot expect to specify the limits of tolerance at different frequencies more closely than with a standard deviation of about ± 2 db.

At the end of the post-exposure tests of electrical output the animal was bled and the circulatory system washed with physiological saline solution. Fixing solution (Heidenhain-Susa) was immediately injected, and the temporal bones removed for further fixation, decalcification and subsequent sectioning. Every fifth section was stained (hematoxylin and eosin) and mounted for microscopic examination. The methods of perfusion, fixation, sectioning, etc. were the same as those used in the earlier phases of this investigation (1). The unexposed ears of 10 of the animals were sectioned to serve as controls of fixation and preparation.

The final histological evaluations of injury were made by Dr. Goldstein and Dr. Davis independently and recorded on a chart similar to that described by Covell and Eldredge (1). Finally more than half of the specimens were reviewed again by Dr. Covell in order to assure that the microscopic criteria and the significance attached to various abnormalities are the same as in the other reports of this series. The microscopic examinations were in general confined to mid-modiolar sections of the cochleas except that other sections were often examined to verify the positions of the holes for the electrodes and to assess the possible local damage done by either the drills or the electrodes.

TABLE I

CALIBRATION OF SOUND SOURCE

Frequency cps	SOUND PRESSURE LEVELS IN DECIBELS								Correction
	1 cc Coupler	Probe Tube at Eardrum, Five Ears					Ave.	Range	
		A	B	C	D	E			
185	118.0	113.9	111.7	113.8	110.1	116.8	113.3	6.7	-5.0
545	124.5	119.0	115.3	119.3	114.1	121.8	117.9	7.7	-6.5
2000	122.0	116.9	112.2	116.4	111.4	117.5	114.9	7.1	-7.0
8000	96.0	112.8	109.2	111.0	108.0	113.8	111.0	5.8	+15.0
	Ave.	115.4	112.1	115.1	110.9	117.5	114.3	6.6	

The reference level is 0.0002 microbar. The sound system was an Atlas PM-25 loudspeaker coupled to an ear-speculum by 1.7 m of garden hose. A constant voltage, 0.10 v, was applied to the terminals of the loudspeaker. The tip of the probe tube was in the external auditory canal within 1.0 mm of the eardrum. The coupler measurements and the corrections to be applied to them have been rounded off to the nearest half decibel. The frequencies are approximate as the oscillator was always tuned to the peak of resonance at or just below the frequency listed.

SECTION II

RESULTS

Absence of the Intra-aural Reflex

A possible disturbing factor in studies of acoustic trauma is the intra-aural reflex. This is the contraction of the stapedius and tensor tympani muscles in response to auditory stimulation. Such contraction is known to cause marked reduction in the sensitivity of the ear to low frequencies (13). We frequently observed spontaneous contractions of the intra-aural muscles in our animals under light surgical anaesthesia, but not when they were deeply anaesthetized. Fortunately the reflex contraction to sound is apparently abolished completely in guinea pigs by even the very lightest dial anaesthesia. We have seen no indication whatever of the intra-aural reflex, even in experiments designed specifically to detect it in animals as lightly anaesthetized as humane considerations would permit. In these experiments we sought for 1) indications of reduction in transmission of low-frequency sound just after the onset of a tone or after superimposing a loud tone-pip on a steady faint test tone; 2) action potentials from fine wire electrodes placed in or on the tensor tympani muscle; and 3) movements of the eardrum, observed through a dissecting microscope. All three methods showed no reflex contractions whatever to sounds of any test frequency (100 to 10,000 cps) at intensities up to 135 db, although very clear effects were seen by all three methods when spontaneous contractions occurred.

Rupture of Eardrum

In only one ear, No. 397, exposed to 185 cps at 148 db (at the eardrum), was the eardrum ruptured. (There may also have been dislocations of the ossicles in this case, but unfortunately the specimen was injured further during the post-mortem dissection.) This ear withstood, with only mild injury to the cochlea, exposures that were far longer than what produced severe injury in ears that did not suffer rupture of the eardrum. No injuries to the drum or to the structures of the middle ear were observed in any of the ears exposed to 545, 2000 or 8000 cps.

Anatomical Injury to the Inner Ear

The anatomical changes immediately following acute acoustic trauma, as seen under the microscope, have already been described by many investigators (1,2,4,14,15,16) and it is unnecessary to reproduce additional photomicrographs to illustrate them. We summarize below the changes that we have found most consistent and that we believe to be useful in predicting whether the ear in question would have suffered from a significant and permanent loss of hearing. The experience of one of us (WPC) with the ears of guinea pigs exposed to intense sound and allowed to recover for periods up to three months before taking the specimens (1) was of great assistance in judging the probable importance of the various changes. All of our present specimens were fixed within three to six hours after the acoustic trauma.

We found no consistent changes in bone, nerves, blood vessels, stria vascularis, spiral ligament, limbus, basilar membrane or tectorial membrane. There were, however, consistent abnormalities, clearly related to the intensity of the exposure tone, in a) the "mesothelial" cells on the tympanic surface (lamella) of the basilar membrane, b) the supporting structures of the organ of Corti, c) Reissner's membrane, and d) the "hair cells" of the organ of Corti. For each of these structures one or more degrees of injury can be specified, as follows:

a) The "mesothelial" cells are completely displaced from the organ of Corti by any exposure sufficient to cause gross rupture or displacement of the organ of Corti. A thinning of the layer with bunching of the remaining cells near the spiral ligament and denuding of small areas under the tunnel of Corti is a rather constant accompaniment of moderate injury to the hair cells, although with longer and somewhat weaker exposures Rüedi and Furrer (16) found degeneration of hair cells with no displacement of mesothelial cells. In itself the displacement of mesothelial cells is probably unimportant and the injury is not permanent, but such displacement is evidence that a given set of changes in the hair cells may be due to acoustic trauma and not to poor fixation and post-mortem degeneration. It also helps to define the area of most violent mechanical agitation of the cochlear partition.

b) The mildest injury to the supporting structures of the organ of Corti is the appearance of vacuoles in or a swelling of the cells of Hensen and/or Claudius and in the supporting cell immediately beneath the inner hair cell. This inner supporting cell seems to be a particularly vulnerable point in the mechanical structure of the organ

of Corti. Sometimes the surface of the cells of Hensen and of Claudius toward the endolymph is irregular and apparently eroded, particularly at the junction between the two sets of cells (see Fig. 1). With slightly greater exposure the

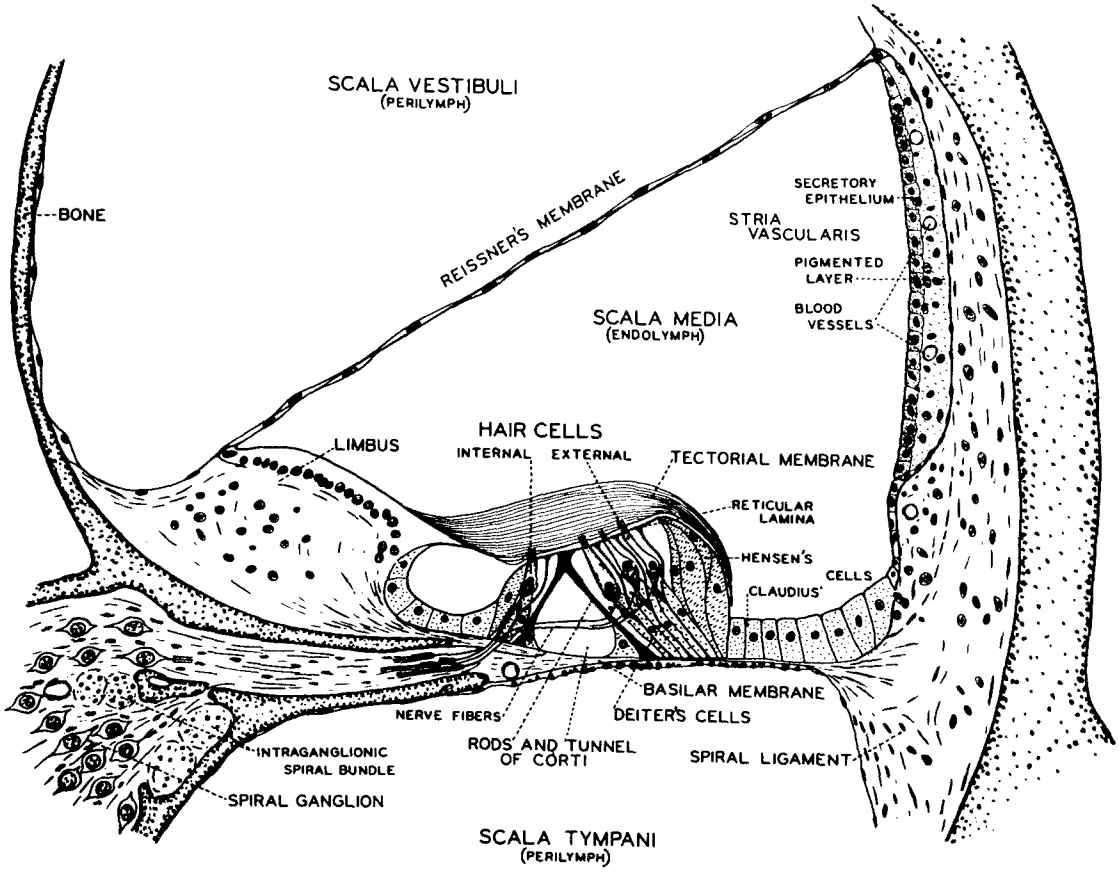

Figure 1. Cross section of cochlear partition of guinea pig in the lower part of the second turn (II 1). The position shown for the tectorial membrane is that given by Hilding (17). It agrees with our own (18) interpretation of microdissections of unfixed specimens and studies under the phase-contrast microscope.

Structures mentioned in the text but not specifically labeled include the space of Nuel (between the outer rod of Corti and the external hair cells) and the "mesothelial cells" on the under (scala tympani) surface of the basilar membrane.

formation of vacuoles may extend more widely to the cells of the external and the internal sulcus. The mere formation of small vacuoles is, however, almost certainly reversible.

As injury becomes more severe the vacuoles become larger, nuclei begin to show darker staining (pyknosis) or dissolution (karyolysis), and cell walls become less clear. These structural changes are often accompanied by the appearance of an amorphous hyalin exudate in the tunnel of Corti, the space of Nuel, etc. In milder cases there may be simply an enlargement of these spaces without visible exudate.

In still more severe injury the cells begin to separate from one another and the supporting cells detach from the basilar membrane. Particularly important, and probably indicating permanent damage, is the separation of the fibers of Deiter's cells from the basilar membrane. Major fissures may also appear along the junction of the cells of Hensen with those of Claudius, particularly if the site of greatest injury is in the second or the third turn of the cochlea.

The severest and most certainly permanent injuries include the complete collapse of the rods of Corti. The inner rods in the apical turn are particularly vulnerable. A more dramatic injury is the complete rupture of the organ of Corti with detachment of Hensen's cells, Deiter's cells, the hair cells, etc. en masse. The most extreme condition is a completely denuded basilar membrane. This condition is much more common, in relation to comparable exposures and to comparable changes in hair cells, in the second and third turns than in the apical and basal turns, perhaps because of the different shape and proportions of the organ of Corti in the different turns.

A relatively rare (four cases) but serious form of mechanical failure of the supporting structures is rupture of the reticular lamina and the extrusion into the scala media of the substance of the hair cells. In general, however, the reticular lamina, like the fibers of Deiter's cells which support it, seems to be a very stiff and rugged structure, stronger than even the rods of Corti. It maintains its shape and integrity even when considerably displaced by the collapse of the inner rods and supporting cells. Sometimes in the basal turn a separation occurs between the reticular lamina and the innermost cell of Hensen that forms the roof of the outer tunnel.

c) A special and rather erratic gross mechanical failure is the rupture of Reissner's membrane. This is easy to see but difficult to evaluate because such a rupture

is too often produced as an artefact of poor fixation, embedding or cutting. We have endeavored to distinguish such artefacts by noting abnormalities of the nuclei and stretching of the cells near the rupture, which usually can be seen in cases of true acoustic injury. Sometimes the rupture seems to have started at the point where the edge of the spiral ligament has been loosened from the bone by the insertion of an electrode or by the breaking off of small bone chips; and a tear in Reissner's membrane, once started, can evidently extend far up and down the cochlea from its point of origin.

d) The hair cells are in general the most vulnerable elements of the organ of Corti. It is well known that the injuries produced by weaker sounds acting over long periods of time are practically confined to these structures and often to the external hair cells alone (16,19). The familiar sequence of changes begins with alterations in the cytoplasm that cause loss of clear detail of the internal structures and some swelling. Small vacuoles may often be seen. Exudate appears in the spaces between the hair cells. The nuclei stain a little more darkly (pyknosis) and are displaced toward the base of the cell. These mild changes we believe are fully reversible.

More severe and probably irreversible injury is indicated by severe pyknosis of the nuclei or fragmentation (karyorhexis) and finally total disappearance of the nuclei. The cytoplasm becomes more swollen and vacuolated and finally the outlines of the cells can no longer be distinguished. When nuclei and/or the cellular outlines have disappeared we feel quite certain that the injury would be permanent.

Evaluation of injury to the hair cells is complicated by two factors. First, these cells are remarkably subject to postmortem degeneration unless fixation by effective perfusion through the blood vessels is carried out immediately after death. The sequence of postmortem changes is almost exactly the same as that caused by acoustic trauma. Apparently even severe and repeated anoxia before death can cause serious degeneration, although more in Deiter's and other supporting cells than in the hair cells (20). Secondly, the external hair cells are almost invariably injured more seriously than the internal hair cells. We get the impression that the acoustic energy is delivered more efficiently to the external hair cells and that their internal structure breaks down first from the overload. The internal hair cell seems better protected, but the internal supporting cell is in a relatively exposed position between the inner edge of the stiff reticular lamina and the

edge of the bony spiral lamina beneath and often is injured more severely than the hair cell adjacent to it.

Evaluation of Anatomical Injury

In order to establish the three-way relationships between anatomical injury, physiological (electrical) failure, and the parameters of the exposure tone we have established a numerical scale of the presumed severity of the anatomical injury. The guiding principle is the probability of recovery of function on the one hand or of permanent hearing loss, either complete or partial, on the other. We have made the hair cells the primary basis of this scale because 1) the hair cells are essential for hearing; 2) the hair cells are highly specialized cells like nerve cells and there is very little chance of regeneration once a hair cell has been destroyed, and 3) the external hair cells are the most delicate indicators of mild degrees of injury. Our estimate must be two-dimensional, however, in that the internal hair cells may be less severely injured than the externals, or neighboring external cells may be injured to various degrees.

Second-order criteria, that make more certain the estimate of permanence and extent of the residual loss of function, are gross injuries to the supporting cells, particularly the detachment of sections of the organ of Corti from the basilar membrane or rupture of Reissner's membrane. Our numerical evaluations are arbitrarily limited to the eight cross-sections of the cochlear duct that are seen in a mid-modiolar section. We therefore give some weight to a severe but localized injury within a quarter or even a half of a turn, whether that injury was produced entirely by sound or whether the operation may have contributed toward it. This allowance is essential in interpreting the changes in electrical output. On the other hand, in assessing the degree of acoustic trauma we must discount possible operative injury.

We believe that rupture of Reissner's membrane is a serious and irreparable injury. It is an unfortunate complication that sometimes the insertion of the electrode seems to have made the cochlear partition more vulnerable to mechanical injury by intense sound, particularly by favoring the rupture of Reissner's membrane, but such accidents have been less frequent with the very small (25 μ) electrodes in our more recent experiments than with the larger (100 μ) wires used earlier.

Our nine-point scale (1, 1+, 2, 2+, 3, 3+, 4, 4+, 5) for expressing the severity of the anatomical injury is summarized in Table II. The major divisions are: 1,

TABLE II

EVALUATION OF ACUTE ACOUSTIC TRAUMA

Rating of Injury	Estimated Severity and Permanence	Anatomic Changes			Electric Loss: Cochlear Microphonic	
					Threshold	Max. Voltage
1	No injury	Entirely normal			0 db	0 db
1+	Certain recovery	Within normal limits			10	5
2	Probable recovery	Hair Cells: Exudate; Nuclei displaced; Mild swelling and pyknosis	Supporting: Erosion and/or small vacuoles	Mesothelial: Some displaced	20	10
2+					30	15
3	Probably permanent partial loss	Severe swelling disintegration pyknosis and/or karyorhexis	Large vacuoles and/or beginning separation of cells	Half gone	40	20
3+						
4	Certain permanent partial loss	Nuclei and/or cell walls absent or clearly destroyed	Deiter's fibers off basilar membrane; Collapse of tunnel of Corti; Rupture of Reissner's membrane	All missing	50	25 and/or badly distorted wave form
4+						
5	Permanent extensive loss	Absent	Separation of organ of Corti from basilar membrane	All missing	60 or more	30 or more

Table II. The anatomical changes refer to specimens fixed within 2 to 5 hours after the acoustic injury. They are graded secondarily by extent as well as by local severity. The condition of the hair cells is the primary basis for rating unless there is gross rupture or collapse of supporting structures (Classes 4 or 5).

The electrical losses are based on measurements made with differential electrodes, but for the two apical turns an electrode at the extreme apex (helicotrema) may be substituted for one in the scala tympani. The measurements were usually made between 10 and 20 minutes after the exposure to intense sound. See text for the test frequencies used for each turn. The electrical indices generally reflect injury only within about 2 mm of a pair of differential electrodes.

"within normal limits"; 2, "probably would recover"; 3, "probably some residual loss"; 4, "certainly some residual loss"; and 5, "residual loss is certain and would be severe." In this table certain electrical criteria are also included. The latter are derived from the experiments to be described below. The foundation of the scale is, however, the anatomical injury, primarily of the hair cells, seen under the microscope and our best estimate of their probable permanence and extent.

Classification According to Anatomical Injury

Not all of the ears used in this study were successfully prepared for histological study. Several pilot experiments were rejected as untrustworthy. In several experiments in which the electrical tests gave unequivocal evidence of very slight or very severe injury the histological work-up was omitted for reasons of economy. A few specimens were spoiled by accidents in fixation, dissection, imbedding or sectioning. Eventually 48 exposed ears, out of a total of 66, and 10 unexposed controls were examined, rated and then classified according to

a) The worst injury seen in any part of the organ of Corti regardless of its position. The approximate location of the center of the region of worst injury was also noted, to the nearest half turn. (Precise localization of the

area of injury was not a special objective of this study. It would require much additional labor because the planes of sectioning, although they usually included the axis of the modiolus, were along different radii for different specimens.)

b) The degree of local injury in the section of the cochlear duct nearest to an electrode. Inasmuch as there was always one pair of differential electrodes near the middle of the basal turn every ear was evaluated for injury to the hair cells near the middle of Turn I. Nearly every ear had one other electrode (or pair of electrodes) in Turn II, III or IV and the corresponding hair cells were also evaluated. In addition any gross injury within half a turn of an electrode was also noted. These evaluations served to define the relation between electrical output and anatomical injury because the pick-up of differential electrodes is localized and therefore the condition of the organ of Corti near the electrodes determines the output. The degree of local and also the worst injury are entered in the Appendix, together with the frequency of the exposure tone, its duration and the acoustic pressure at the eardrum.

The Location of Worst Injury

The injuries center in different turns, depending on the frequency of the exposure tone, roughly as follows:

185 cps, at the junction of Turns III and IV
545 cps, in the lower half of Turn III
2000 cps, in the lower half of Turn II
8000 cps, in the upper third of Turn I

The areas of injury for the two middle frequencies center quite close to the usual positions of the electrodes in Turns II and III respectively. This was expected because 545 and 2000 cps are almost exactly the frequencies for which the phase shift is π in Turn III and Turn II respectively. The frequency for "π phase shift" is a little higher than the critical "2π frequency" below which the amplitude of the cochlear microphonic falls off sharply (8). (Figure 13 in reference 8 shows the relations between these frequencies and position along the cochlear partition.) The usual center of injury for 185 cps is a little more basal and for 8000 cps it tends to be a little more apical than would be expected from the chart in reference 8. This recalls the similar tendency reported by Lurie, Davis and Hawkins (2) for acoustic injury to occur closer than expected to the center of the basilar membrane. In the present experiments, however, the positions and extent of injury have not been precisely mapped and we note them chiefly to show

that the frequencies of the exposure tones were fortunately chosen. They actually produced injuries that usually centered quite close to the positions at which the electrodes sampled the electrical output of the organ of Corti. The position of the injury was not determined by the presence of the electrodes, however, except in perhaps three or four instances in which the bone chipped badly. The positions were in general the same whether or not electrodes had been inserted in a particular experiment.

In an earlier section we noted that the type of gross injury to the supporting structures varied according to the part of the cochlea that was injured. Collapse of the tunnel of Corti is most frequent at the apex, for example, and rupture at the Hensen-Claudius junction in the second and third turn. These same differences are, of course, correlated also with the frequency of the exposure tone because the position of the injury is closely correlated with frequency. We cannot say whether it is the frequency of the exposure tone or the details of structure or both that determine the way in which the organ of Corti breaks down. Perhaps when the effects of variations in intensity and duration have been investigated an analysis of the mechanics of the injuries will become profitable.

Electrical Indices of Injury

Some indices that might be derived from our measurements of thresholds and of voltages are

1) Wave form and amplitude of cochlear microphonic during the exposure.

2) "Threshold" of action potentials.

3) Maximum voltage of action potentials.

4) "Threshold" of the cochlear microphonic.

5) Maximum voltage of cochlear microphonics.

6) Strength of stimulus at which the maximum voltage (CM or AP) is reached.

7) Recovery of Items 2 through 6 during one or two hours after the first test.

On the basis of many experiments reported elsewhere (11) and study of our present data we offer the following critical evaluation of these various indices. They may be useful in an interpretation of previous studies (21,22,23) and for the

planning of future experiments that involve the electrical output of the cochlea. Unfortunately most of the indices are either untrustworthy, limited in scope, or redundant.

1) Wave form and amplitude of the cochlear microphonic during exposure were recorded photographically and monitored acoustically, but the photographs have not been analyzed because of the complexities of the non-linear behavior of the ear at high intensities. The most useful information yielded by these observations was that a reduction in CM voltage and changes in wave form often occurred quite suddenly and rapidly after a few seconds of exposure with relatively slow further shrinkage thereafter. Subharmonic frequencies and the irregular "crackling" described by von Gierke (24) were regularly seen and heard during exposures to 8000 cps but never at 2000, 545 or 185 cps.

Distortions of wave form were also observed during many tests after exposure. They frequently served as the basis for identification of the residual response as AP or perhaps "summating potential" instead of CM. This distinction was often essential for correct estimation of the threshold of the microphonics and of their maximum voltage.

2) We expected that the threshold of action potentials would be a very useful index as it is obviously most closely related to subjective and behavioral thresholds. The action potentials can easily be measured, using tone-pips as stimuli and canceling the CM. A pair of differential electrodes in parallel act as one electrode: the other is a reference electrode on the neck. This combination effectively records the action potentials of the nerve impulses as they pass through the modiolus (10). It thus detects any activity in the auditory nerve, regardless of which turn of the cochlea contains the electrodes. (We regularly employed the basal turn.) But it is not possible to record the action potentials selectively from local areas of the organ of Corti as we do with the cochlear microphonic. This important difference indirectly imposes a serious limitation on the information yielded by the threshold for AP in an injured ear. In the first place, tone-pips must be used as stimuli in order to set up well-synchronized volleys of nerve impulses. But tone-pips contain many frequencies other than the central frequency, and although their intensity level is far enough below that of the central frequency to make them relatively unimportant for the normal ear (25) they soon become the effective stimulus for an ear that has been injured locally. The change in threshold may then measure the physical properties of the stimulus as much as the extent of injury to the ear. The presence of action potentials simply means qualitatively

that some part of the cochlea and auditory nerve is still able to respond more or less normally.

In the second place, although high frequencies such as 8000 cps activate only the basal turn, low frequencies like 545 and 185 cps spread along the entire basilar membrane. The basal turn yields a good CM response to a 500 cps tone-pip, and apparently a good nerve response also. The threshold for AP (500 cps tone-pip) may be elevated only 20 db by exposure to 545 cps although severe anatomical injury with practically complete elimination of CM may have been produced in the two apical turns. We conclude that the threshold of action potentials is not a valid measure of the severity of local injury in the cochlea except perhaps for the extreme basal end and for frequencies near the upper limit of the animal's hearing. A similar conclusion can be drawn from the studies of Schuknecht and Neff (26) who correlated the conditional reflex thresholds of cats with surgical injuries of the apex of the cochlea. Destruction of the apical turn might elevate the animals' thresholds for low tones by about 40 db but a complete low-tone deafness was never produced.

3) A further drawback to the use of either the threshold or the maximum voltage of action potentials is their great lability. The voltage is reduced and the threshold elevated by intensities and durations of exposure that do not cause clear anatomical injury. Furthermore the recovery following an exposure may be considerable and rapid so that it is difficult to specify at what interval after the exposure the action potentials should be measured. All of the indices that involve the action potentials seem to be much more closely related to the problem of auditory fatigue and reversible hearing loss than to our present problem of anatomical injury and permanent hearing loss. We have therefore discarded the action potential as the basis of useful quantitative indices of injury.

4) The threshold of the cochlear microphonic (CM) is an informative and reliable index. We believe that it is a logical index as we think that CM is the immediate stimulus to the nerve endings (10,27). As an index of injury, however, it is subject to several important requirements. First, in order to gain information as to local conditions in a given part of the cochlea, it is necessary to introduce differential electrodes into that region, one in scala vestibuli and one in scala tympani or at the helicotrema. A single electrode in scala vestibuli with a distant reference electrode gives much less precise information. An electrode on the round window with a reference

electrode on the neck gives fairly good information as to the lower half of the basal turn but not beyond. The basic advantages of the differential electrodes are that they exclude the action potential and they detect CM effectively from only one or two millimeters along the cochlear partition. This localized pick-up, in contrast to the generalized pick-up of AP, makes it possible to detect a localized injury.

Second, test tones of different frequencies do not test different parts of the cochlea. All responses originate in the area near the differential electrodes. This proposition we confirmed experimentally many times (7). For example, electrodes in the basal turn regularly yielded good thresholds for all frequencies from 8000 to 150 cps. The thresholds might be elevated by 30 to 50 db following acoustic trauma but the elevations were always the same within 4 or 5 db, i.e. within the errors of measurement, for all test frequencies that gave a good initial response. The anatomical injury was, however, quite different in different parts of the cochlea. (The correspondence between the average change of threshold of CM and the local anatomical injury will be described in full below.)

Third, we believe that most experiments that have used CM as an index of injury suffer from failure to recognize adequately that the usual placement of only one electrode on the cochlea, either on round window, on the apex or in a hole, gives a partially but not completely selective pick-up from which no clear conclusions can be drawn either as to local conditions or as to the over-all performance. With a distant reference electrode changing the frequency only partially clarifies the picture. A high frequency tests the basal turn only. A low frequency activates all parts of the cochlea, but if the selective electrode is on the round window the output is still dominated by the basal turn. Local injury in the basal turn cannot easily be measured, however, because the more apical portions do contribute somewhat to the recorded potential, but to an unknown extent. Of course, this criticism does not apply to the study of conditions like anoxia (22, 28, 29) that presumably affect all parts of the cochlea alike. In such experiments, however, the use of many different test frequencies is superfluous.

Fourth, even with differential electrodes the exclusion of small contributions of CM from distant parts of the cochlea is not quite complete. If the threshold for local response has been elevated by 60 db the response from a still normal but remote part of the cochlea or a trace of AP response may be detected and will mask the true local threshold. We cannot distinguish reliably between

a 60 db elevation of threshold and complete abolition of local response.

Fifth, after severe acoustic injury small potentials of unknown significance (30,31) may still remain. They may or may not show "rectified" or other nonsinusoidal wave forms. These may be very difficult to distinguish from the "true" CM that is sensitive to anoxia. They are the "second-order microphonics" postulated by several authors (28,11) and perhaps also the second-order summating potentials (S_2 and S_3) described by Goldstein (31). They make it difficult to diagnose a complete local destruction of the organ of Corti.

Finally, there is sometimes a partial recovery or else further deterioration of CM responses after injury. The changes are not so rapid or so large as for AP but they require that the interval between trauma and test be standardized. (Our average interval for the first post-traumatic test has been about 15 minutes.)

5) The maximum voltage of the CM is also a useful index. It is easier to measure objectively than the "threshold." In connection with measurements of threshold it permits some analysis of the nature of injury. If our hypothesis (11,27) as to the generation of the microphonic is correct, a change in the maximum voltage indicates a reduction in the DC voltage of the postulated "battery." An elevation in threshold equal, in decibels, to the reduction in maximum voltage is also to be expected because of the linear relation between sound intensity and voltage of CM near threshold. (See Fig. 2.) But if the threshold is elevated more than the maximum voltage is reduced it indicates an additional defect. Our theory assumes that the flow of current from the "battery" is modulated by a variation in series resistance that is proportional (within limits) to the mechanical displacement caused by the sound waves. The variable resistance is tentatively located in the upper end of the hair cells. An "additional defect" that would change the threshold for CM without reducing the maximum CM voltage might be a reduction in the change of resistance that is caused by a given displacement of the hairs. It might also be any conductive defect, including a separation of the hairs from the tectorial membrane, that prevents a given sound pressure from causing the normal amount of movement of the hairs.

The suggestion by Tonndorf and Brogan (22) that maximum CM measures the DC voltage of the cochlea obviously anticipates part of this hypothesis. Regardless of theoretical interpretation we shall show below that changes in the

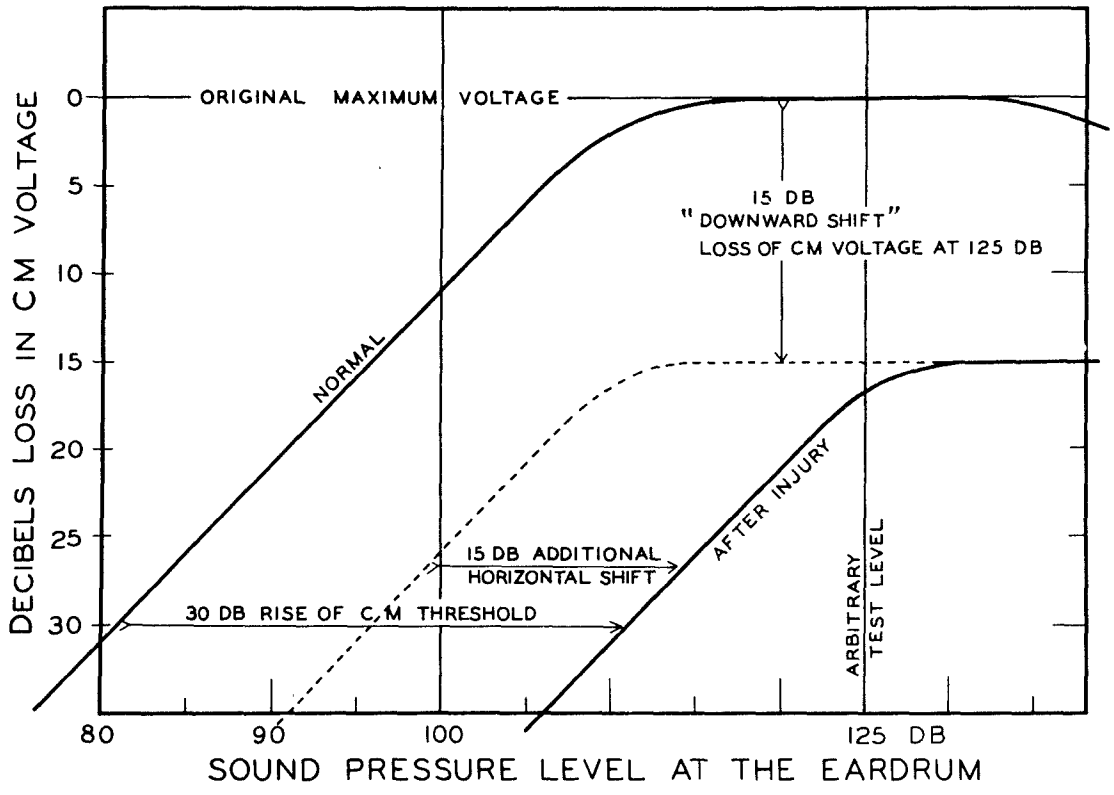

Figure 2. Input-output curves of CM voltage to show the double shift, downward and to the right, that is typically produced by acoustic injury. The electrical measurements in this case would show a 30 db elevation of CM threshold and a 16 db loss in CM voltage at 125 db sound-pressure level. These reductions correspond to a moderate but probably reversible degree of injury (2+). These particular curves represent, for convenience, a rather insensitive animal. The sloping segments usually lie about 15 db farther to the left.

maximum voltage correlate with the observed anatomical changes at least as well as do the changes in threshold.

Practical limitations on the measurement of the maximum

CM voltage are the following:

a) At frequencies below 1000 cps the wave form becomes distorted by harmonics before the maximum voltage is reached. This makes it impossible to measure the maximum of the fundamental accurately without a wave-analyzer.

b) At frequencies above 2000 cps the voltage of CM goes through a maximum, as described in detail by Wever and Lawrence (28). The maximum is often unstable and the voltage falls appreciably if stimulation is continued. It seems that some "fatigue" effect obscures the true maximum.

c) After acoustic trauma a higher sound level is needed to elicit the maximum voltage of CM, sometimes so high that there is possibility of further acoustic trauma if the necessary sound level is applied.

For these reasons we confined our measurements of maximum CM output to the frequency 2000 cps for the basal and the second turn, and to 500 cps (in spite of some distortion) for the third turn. (Tones of 2000 cps do not stimulate the third and fourth turn (7).) We have not made satisfactory measurements of maximum voltage from differential electrodes in the apical turn. Furthermore we arbitrarily limited the intensity level of our tests to about 125 db (re 0.0002 microbar, at the eardrum) which is about 20 to 25 db above the level at which maximum CM output is normally reached. The actual measure therefore became the voltage of CM at 2000 (or 500) cps at this arbitrary high intensity. This output was the maximum output before acoustic trauma and it was either maximum or near maximum afterward unless the trauma had been very severe indeed.

For the same three reasons it was not practical, after exposure, to measure the strength of stimulus at which the maximum CM voltage was reached. The same information that this measurement would yield can be deduced from the change in threshold of CM and the reduction (in db) of the maximum output, provided we are willing to make the reasonable assumption that the shape of the input-output curves remains the same although they may be shifted either downward (reduction in maximum CM voltage available) or horizontally to the right (loss of sensitivity of the "transformer"). The actual results, diagrammed in Fig. 2, show that both of these shifts actually occur.

6) Recovery of CM thresholds and of maximum outputs was measured after one and sometimes also after two hours. When recovery occurred it was usually less than the recovery

for AP. As a practical index of injury the recovery of CM proved to be redundant because it correlated very closely with the initial rise of threshold measured about 15 minutes after exposure. With slight losses the recovery was considerable while with severe initial losses (over 40 db) the threshold remained stable or rose still further. The later measurements were often complicated by the presence of some fluid, either serum or perilymph, in the bulla or by the manipulations necessary to remove it. We have therefore not analyzed in detail our data on recovery of CM threshold or voltage.

Correlation of Electrical Indices with Local Pathological Changes

The two electrical indices finally selected as the most reliable and informative were 1) the elevation of threshold for CM and 2) the reduction in the voltage of CM at 125 db. For all 48 ears in which adequate anatomical preparations were available the decibel losses were plotted against the pathological classification of the hair cells. The correlations were encouraging, but it was immediately obvious that the electrical output was often greatly reduced, even with rather mild injury to hair cells, if there was a gross rupture of either the organ of Corti or of Reissner's membrane elsewhere in the same turn. Sometimes an injury even two turns away had a severe effect, although in animals 390, 350 and 394 such distant injuries caused little depression. Mere incipient rupture of the organ of Corti seemed not to be very important.

The correspondence between visible injury to the hair cells, with allowance for an invisible depressing action from gross nearby injury, is a little more consistent for the loss of CM voltage at 125 db than for the elevation of threshold. Unfortunately we did not obtain measurements of the maximum voltage at all of the proper frequencies in the early experiments, i.e. at 2000 cps for Turns I and II, at 500 cps at Turn III, and at 250 cps at Turn IV. The distribution for Turn I is shown in Fig. 3. (The number of technically satisfactory and comparable cases for the other turns is too small to warrant graphic presentation. The data are in the Appendix. Nearly all cases for Turns II, III and IV show a loss of CM voltage of more than 30 db and probably permanent changes in the hair cells and/or gross injury to one of the supporting structures.)

Figure 3 shows a fairly clear division at 10 db loss between the ears that retained a very high proportion of normal hair cells and those with a predominance of abnormal (but probably still viable) hair cells. It is also clear

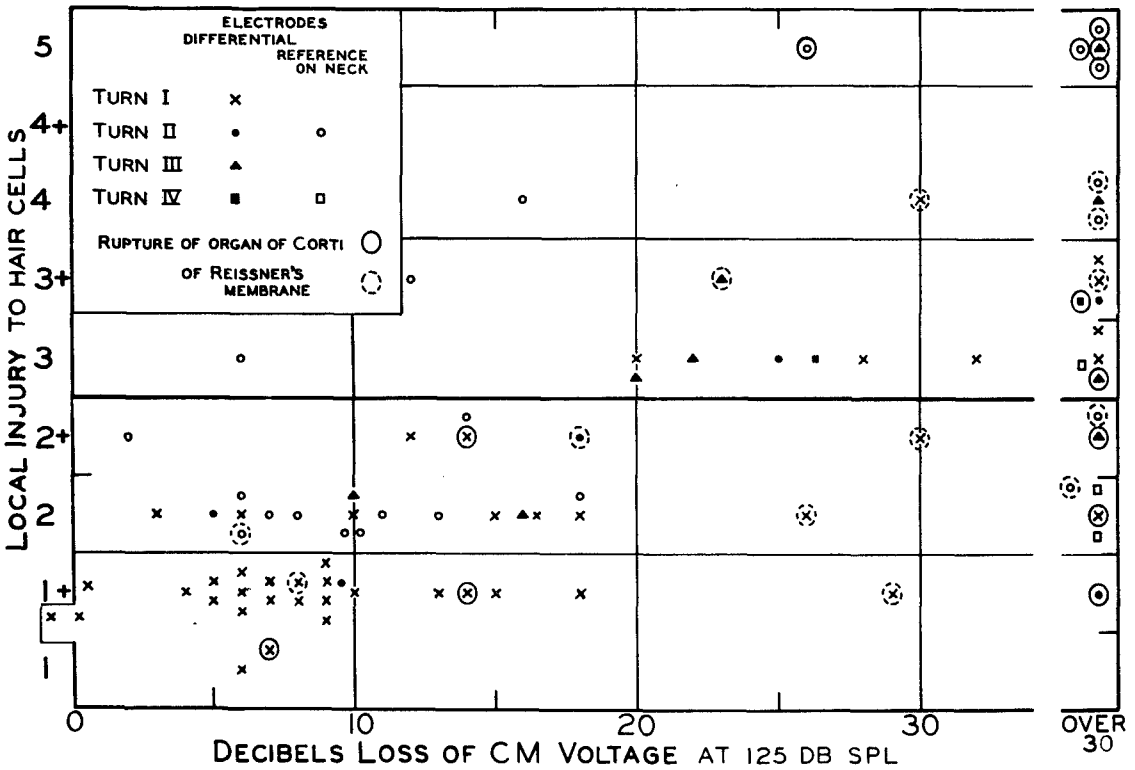

Figure 3. The losses in maximum voltage of the cochlear microphonic measured with differential electrodes are plotted with crosses or solid symbols and those with a reference electrode on the neck with open symbols. Note that the latter from Turn II (circles) tend to lie to the left of the solid symbols and those from Turn IV (squares) to the right. The solid symbols and crosses group more consistently along the diagonal of the chart. The large losses of voltage that were measured as simply "more than 30 db" are grouped arbitrarily in the column at the extreme right. The large electrical losses with slight injury to hair cells (lower right corner) are explained by ruptures of organ of Corti (solid encirclement) or of Reissner's membrane or operative injury (broken encirclement). Only ruptures and injuries within one turn of the electrodes are considered. The heavy horizontal line divides the probably permanent injuries to hair cells (above) from the probably reversible injuries (below). All encirclements represent probably permanent injuries.

that losses of 20 db or more are associated with the "probably permanent" injuries, and that a loss of 30 db or more is in every case associated with severe injury to either the hair cells, the supporting structures, or both.

The corresponding relationships appear almost equally well for the elevation of threshold of CM (Figs. 4 and 5)

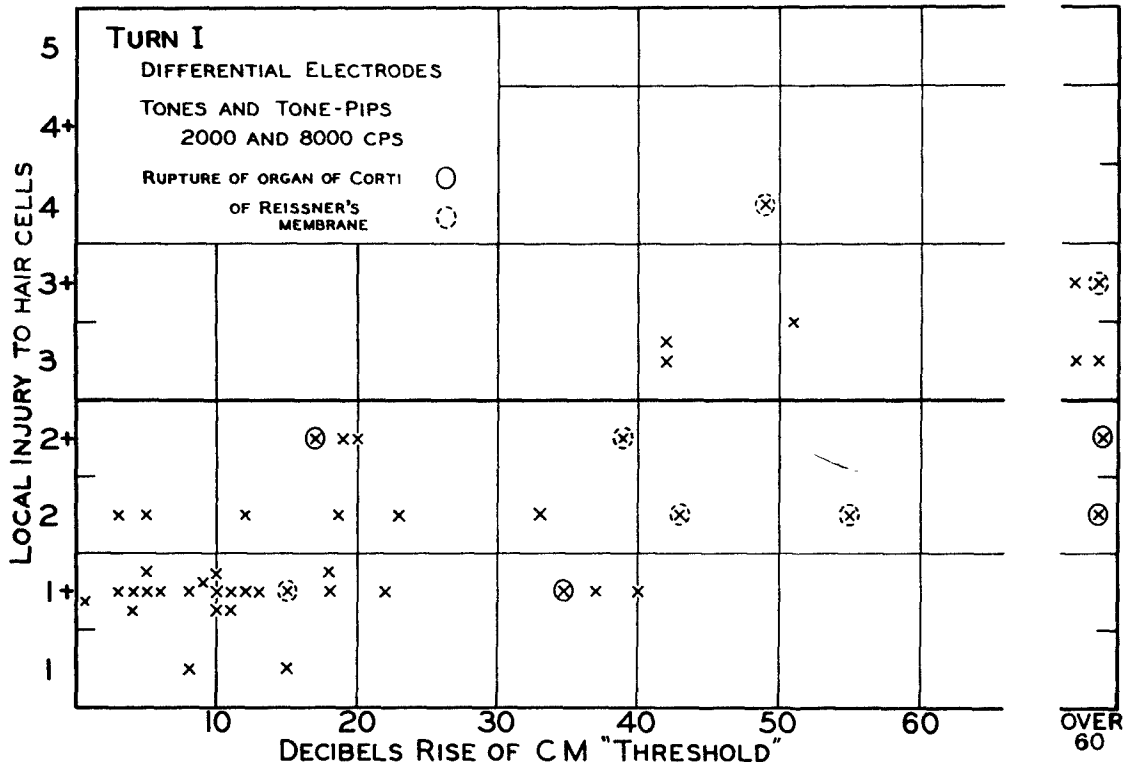

Figure 4

but the numerical values that divide the three major grades of injury are higher. It is 20 db elevation of threshold, not 10, that seems to mark the presence of definite but probably reversible injury; 20 db indicates probably permanent injury and 60 db indicates severe and certainly permanent injury. Each of these numbers happens to be just double the corresponding number for the diminution of CM output at 125 db.

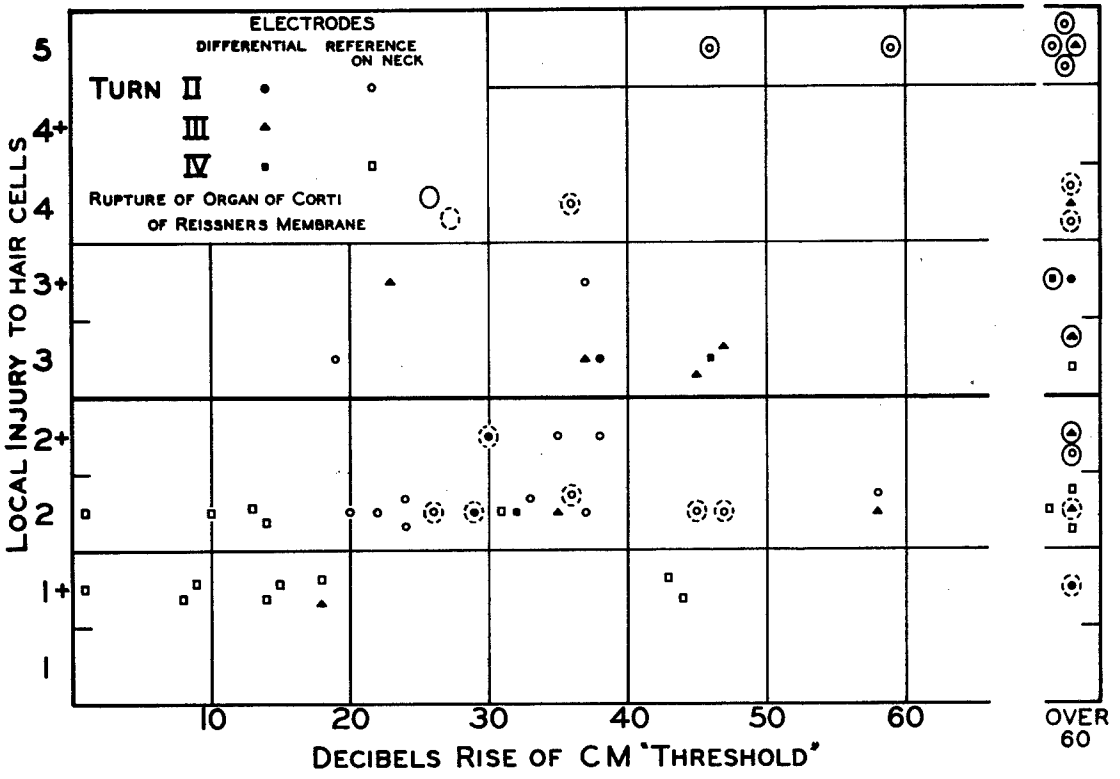

Figure 5

Figures 4 and 5. In these two figures are plotted the elevations of the "threshold" of the cochlear microphonic as measured in Turn I (Fig. 4) by differential electrodes and in the other turns (Fig. 5) with both types of electrode. The same symbols are used as in Fig. 3. Crosses and solid symbols represent differential electrodes. Note that the decibel scale (abscissa) is compressed by a factor of two relative to Fig. 3. Figure 5 shows that differential electrodes give the most consistent relationship between electrical data and anatomical injury. There is little or no relation between injury to hair cells and the "threshold" of CM in Turn IV (open squares) when the "threshold" is measured with a reference electrode on the neck. Even for differential electrodes the data scatter more than in Fig. 3. Note the aberrant points for Turn III (solid triangles) in Fig. 5. The corresponding points based on maximum voltage are in line in Fig. 3. See text for the test frequencies employed for Turns II, III and IV.

The elevation of CM threshold and the reduction of CM voltage at 125 db are highly correlated with one another. The relationship is shown graphically in Figs. 6 and 7.

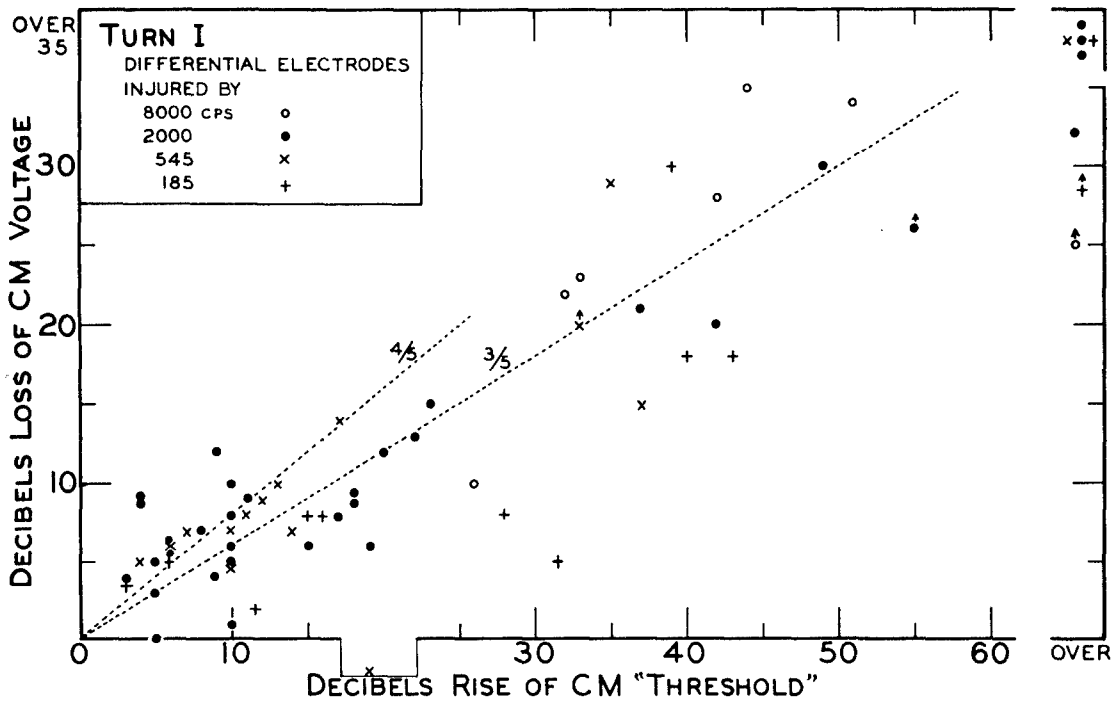

Figure 6

Here it is evident that the two-to-one relationship noted in the previous paragraph is an oversimplification. The ratio for Turn I is about 5 to 3 while for the upper turns it is more nearly 5 to 2. The scatter is so great that we prefer not to emphasize this difference between the basal and the higher turns until we have collected more data with special attention to the CM output at 125 db and its possible contamination by AP or SP. The approximate 2 to 1 ratio between elevation of threshold and depression of "maximum voltage" is clear, however, and is of considerable theoretical importance.

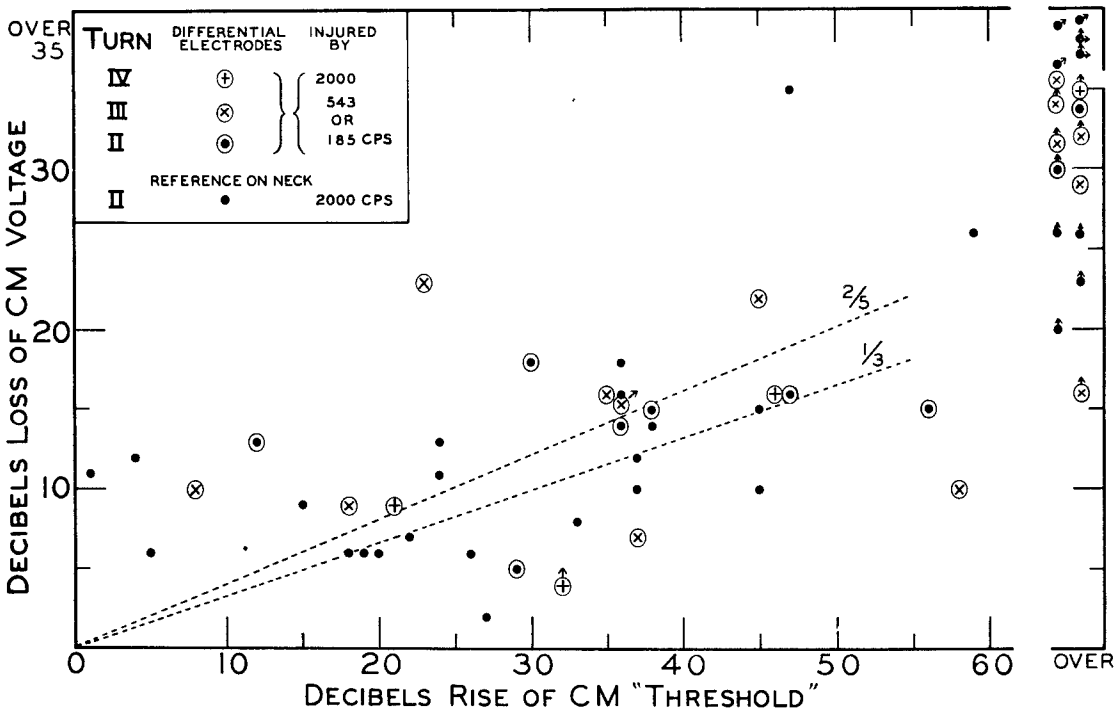

Figure 7

Figures 6 and 7. The reduction in maximum CM voltage is fairly well correlated with the elevation of threshold in Turn I (differential electrode). The correlation is not so good for the upper turns, particularly when a single electrode is used with a reference electrode on the neck. The dotted lines are not fitted to the data but are merely drawn to illustrate slopes of 4 to 5, 3 to 5, 2 to 5 and 1 to 3 respectively. Small arrows attached to symbols in this and other figures indicate that the point represents a minimum value and that the true value is probably somewhere in the direction indicated.

Schematic input-output curves for CM before and after a fairly severe injury are shown in Fig. 2. The injury has shifted the curve downward by 15 db, but we cannot explain the additional horizontal shift simply as a result of "weakening the DC battery that provides the energy for the cochlear microphonic." (11, 22, 27) We must assume a second injury also: some elevation of "threshold" of the mechanism that modulates the DC energy to generate the AC microphonic, or a loss of efficiency in delivering the mechanical energy to the critical spot. We may speculate that the amount of electrical energy available is rather closely related to the visible pathological changes. The change in threshold of CM may be related to the visible changes in two different ways, or to the visible changes and also to a set of invisible changes. In each case the total elevation of threshold should be greater than the loss of maximum voltage and also more variable.

For the next step in our analysis we have therefore accepted the reduction in CM voltage at 125 db as the electrical index of choice to evaluate the injury in the ears for which anatomical specimens are not available. Fortunately, as Fig. 3 shows, the relation of "CM max" to anatomical injury is reasonably good for a single exploratory electrode in Turns II, III and IV as well as for differential electrodes. When the CM voltage at 125 db was not determined at an appropriate frequency or when distortion of the wave form was very severe (as in studies of Turn IV) we used the shift in threshold of CM as the most valid available index of injury.

Relation of Injury to the Frequency of the Exposure Tone

It is well known (2) that the severity of acute acoustic trauma depends on the duration as well as on the intensity of the exposure tone. Also high frequencies seem to be more injurious than low frequencies (32). The present series of tests was planned to explore primarily this dependence of injury on frequency for the middle and lower parts of the auditory spectrum, and secondarily the relation between intensity and duration. The duration most frequently employed was one minute with extremes of five seconds and six minutes. The intensities were chosen, as the tests proceeded and on the basis of the electrical tests alone, to produce what we tentatively believed would be "probably permanent" injury. It is of some interest that even before the anatomical specimens were available we had learned to recognize an elevation of 60 db in threshold as the criterion of "very severe" injury and an elevation of 40 db as the basic indication of "probably permanent" injury, and 20 db as "mild but significant" injury. (These tentative criteria based on elevation of

"threshold" proved to be correct, although the "maximum CM voltage" seems to be a still better index and we have discarded our other tentative criteria such as the recovery of action potentials.)

The Appendix shows for 66 ears the frequencies, durations and intensities of the various exposures and also our best assessment of the severity of the most serious injury in any part of that cochlea. The microscopic appearance of hair cells and supporting structures, if available, was the basis of the assessment; otherwise the loss in CM output at 125 db or finally the rise of threshold for CM. In the latter cases the electrical changes at the set of electrodes showing the greatest change were used as guides.

In Fig. 8a, b, c, d is plotted the position of each

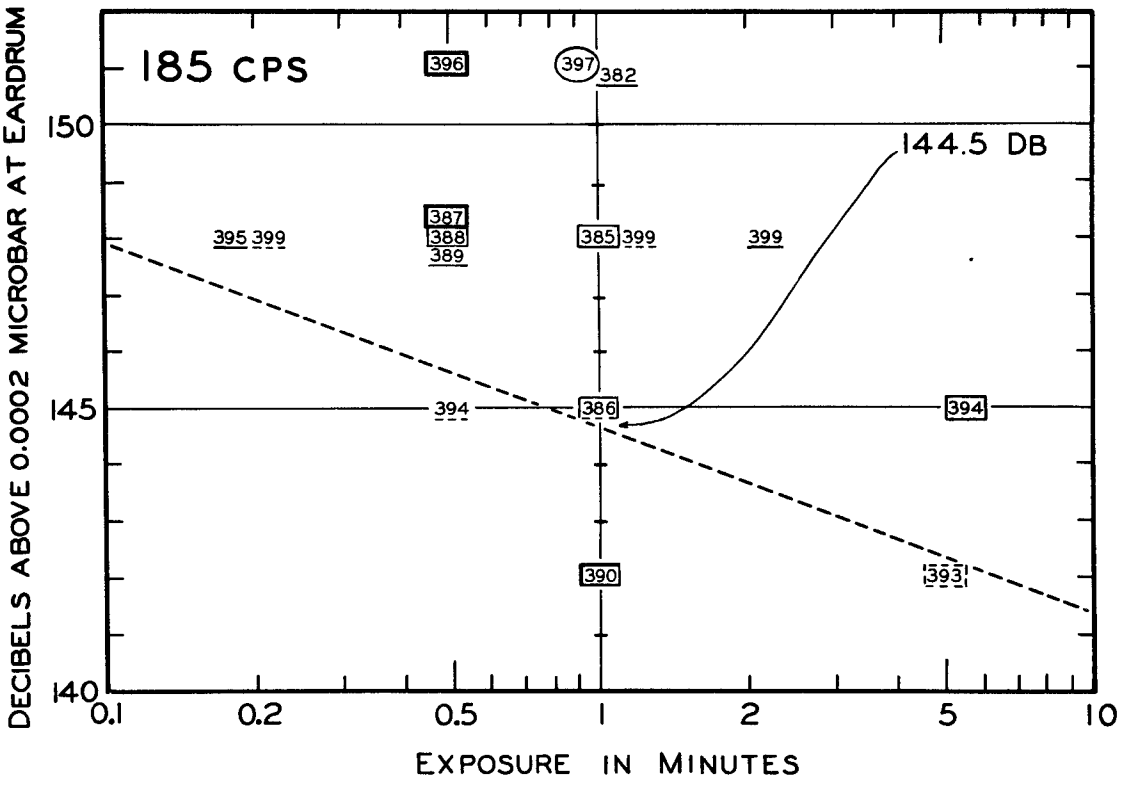

Figure 8a

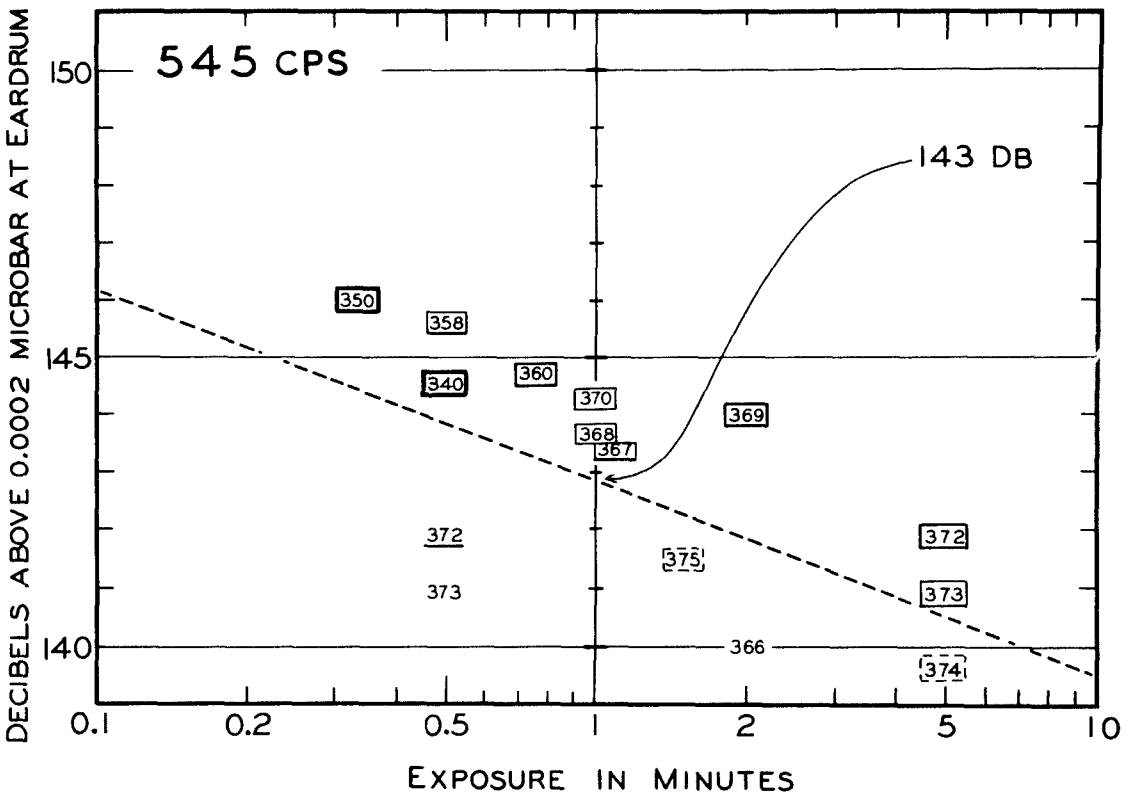

Figure 8b

Figure 8. The severity of injury is indicated by the boxes or underlining as shown in 8d. Complete boxes indicate that microscopic evidence was available. The underline gives the severity of injury judged from electrical evidence alone. The heaviness of the lines indicates the most severe injury seen in any part of the cochlea or inferred from the greatest electrical losses at any pair of electrodes. The numerals give the number of the animal. They are located vertically according to our best estimate of the intensity of the exposure except where two or more boxes would overlap. The diagonal lines are located by eye to separate as consistently as possible the "permanent" from

the "temporary" injuries, with the restriction that the lines for all four frequencies are straight lines with the same slope. The slope chosen is 1.0 db per double of duration. The dotted line is the corresponding dividing line given by Fernández, von Gierke, Eldredge and Davis (9), for exposures to 9000 cps. It was located on the intensity axis by applying our correction factor derived for coupler measurements at 8000 cps to the measurements of field pressure at 9000 cps that were reported by these authors. The location of the dotted line is therefore only approximate.

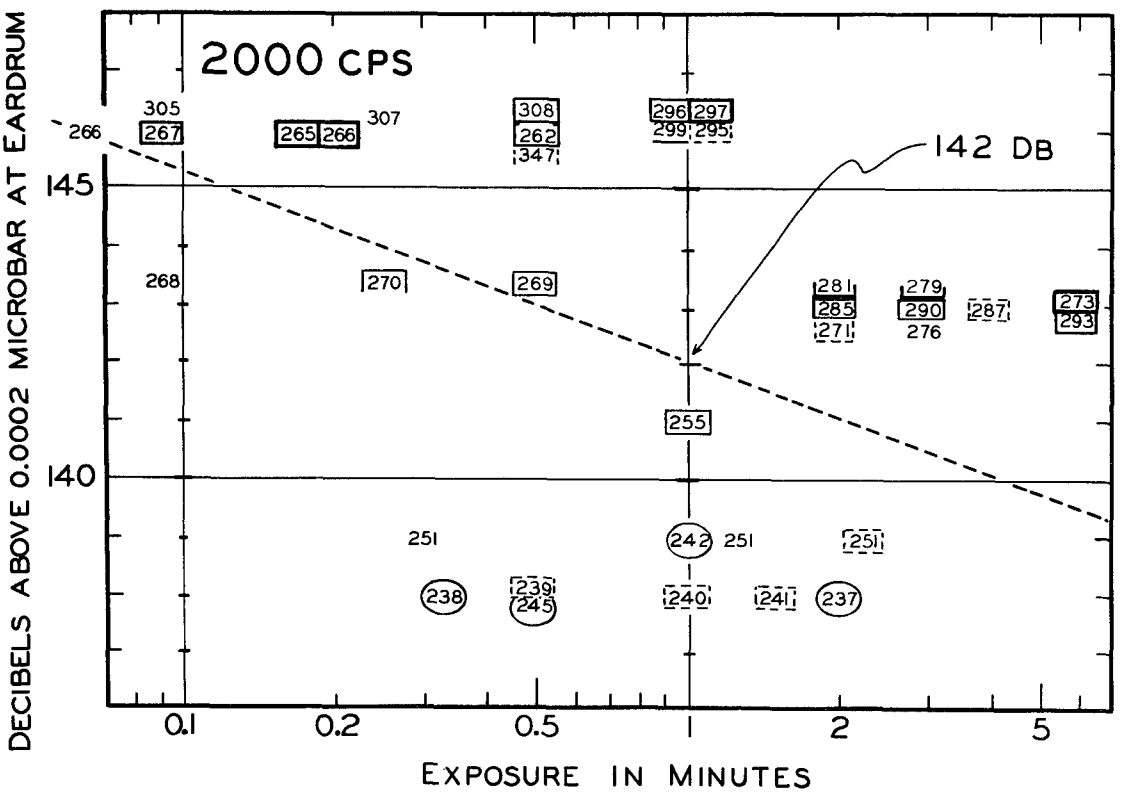

Figure 8c

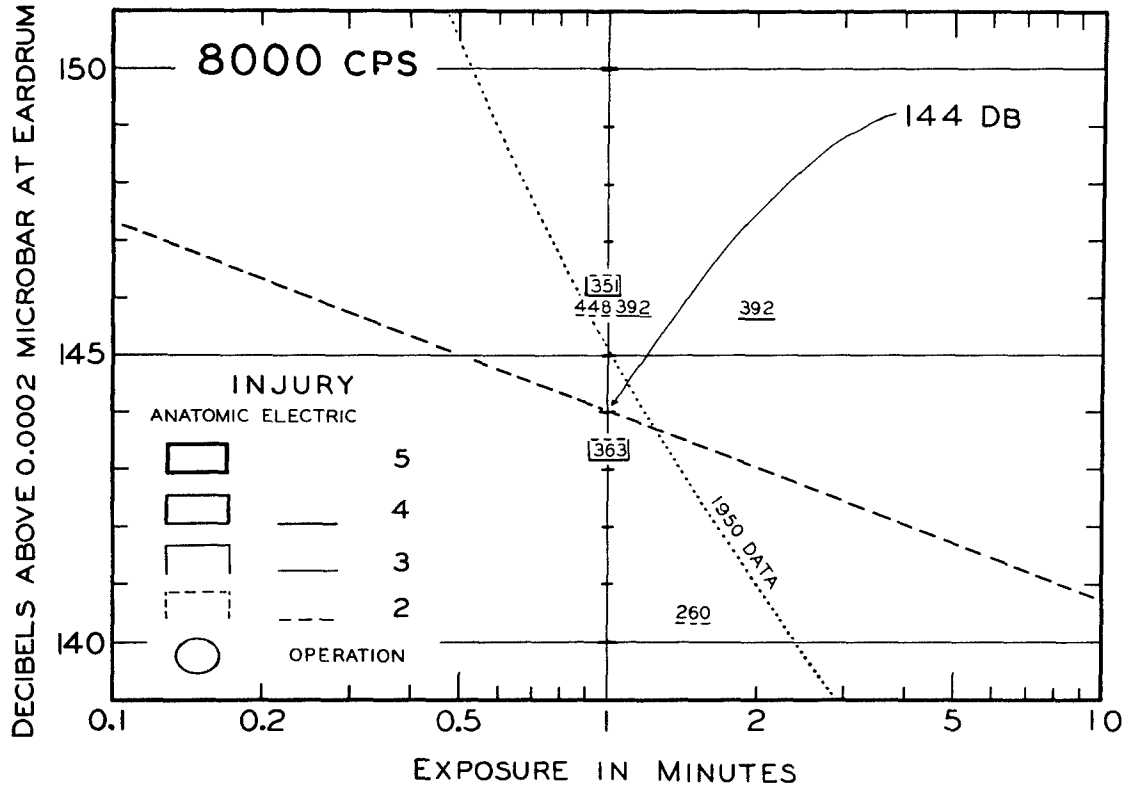

Figure 8d

ear according to the frequency, intensity and duration of its exposure. The numbers for ears that were "severely and permanently" injured (grades 4, 4+ and 5) are boxed with heavy lines. Those that were "probably permanently" injured (grades 3 and 3+) are lightly boxed. The ears for which the evidence of severe injury is purely electrical are incompletely boxed. A few cases which are complicated by apparent injury from the operation or by rupture of the eardrum are encircled, and these complicated cases are discounted in our interpretations.

Figure 8b, for 543 cps, shows the clearest division between "probably reversible" and "probably permanent" injury. All of the latter and none of the former cases lie

above and to the right of the dividing line. This line passes very close to 143 db at the one-minute duration. It slopes downward at a rate of approximately 1 db per doubling of the duration of exposure. The location of this line seems well established within ±1 db.

The division is not quite so clear for 185 cps (Fig. 8a). The electrical evidence for the first two exposures of animal No. 399 showed less injury than would be expected while another animal, No. 390, suffered severe injury from a much milder exposure. We have drawn a "dividing line" which passes through 144.5 db at one-minute duration. There were not enough mild exposures to establish this line clearly and it may be in error by ± 2 db.

For 2000 cps (Fig. 8c) we have more cases and a wide range of durations but also more variability. Even after discounting the four cases that seem to have been injured by the operation as well as by sound there is considerable intermingling of severe and moderate injuries. We believe that this represents true biological variability. We locate our "dividing line" at about 142 db, but with an uncertainty of about ± 2 db. (This "uncertainty" is merely a subjective estimate and not a calculated standard deviation.)

The number of cases at 8000 cps (Fig. 8d) is very small but it gives us the important information that the dividing line lies at about 144 db with an uncertainty of at least ± 2 db. The injury to the hair cells tended to be very "spotty" in these animals, but the intensity required seems clearly to be about the same as for the three lower frequencies.

In Fig. 8d we have compared our present results with the preliminary results reported by Fernández, von Gierke, Eldredge and Davis (9). The dotted line represents a segment of their dividing line between exposures to tones of 9000 cps that produced "mild" or "severe" injury. Only electrical criteria of injury were used in that series. The location of the (dotted) line on the intensity axis is only approximate as we have assumed that the correction for resonance in the ear canal is the same at 9000 cps relative to a field measurement as it is at 8000 cps relative to a coupler measurement. We should expect a correction of this order of magnitude because the human ear shows an increase of pressure at the eardrum relative to the field pressure at the entrance to the canal of 12 db at 4000 cps. The guinea pig ear canal is smaller and should have a similar resonant peak at a higher frequency.

Our information relating injury to frequency is collected in Table 3 and can be summarized as follows: a probably permanent injury is produced in the ear of an anesthetized guinea pig by a one-minute exposure to a tone of 143 db (± about 2 db), measured at the eardrum and independent of frequency from 185 to 9000 cps. The scanty present data suggest that in the range from 10 seconds to five minutes the injury is more strongly related to the energy level than to the duration.

TABLE III

FREQUENCY VS PROBABLY PERMANENT INJURY
BY ONE-MINUTE EXPOSURE

Cps	Db at Eardrum	Uncertainty	Site of Worst Injury
185	144.5	± 2 db. Not enough variety of exposure.	Turns III and IV
545	143	± 1 db. Very consistent.	Turn III, lower half
2000	142	± 2 db. Many individual differences.	Turn II, lower half
8000	144	± 2 db. Few cases. Injury is "spotty."	Turn I, middle and upper thirds.
Ave.	143.5:	No trend with frequency.	

The decibel values are derived from Fig. 8. Each is the intercept of the best dividing line between "probable recovery" and "probably permanent injury" with the ordinate at one-minute duration of exposure.

SECTION III

DISCUSSION

The independence of injurious effect from frequency is at first surprising, but it clearly is due to the unexpectedly high pressures measured at the eardrum at 8000 cps. Here the pressure is 15 db higher than the coupler measurement for the same voltage applied to the loudspeaker. It is also above the field measurement reported by Fernandez, von Gierke, Eldredge and Davis (9) as producing a similar injurious effect. The higher pressure at the eardrum is probably caused by the resonance of the external ear canal. Further comparisons between the pressures at the eardrums of guinea pigs and of man in sound fields of knownintensities will be reported at a later date. We can then properly consider the relation of our traumatic acoustic level of 143 db at the eardrum to previous studies on man and on guinea pigs (1,2,3,4,9,30).

In any extrapolation of our present data to the problem of acoustic trauma in man we can probably assume with some confidence that the organ of Corti of man has about the same mechanical strength that it has in the guinea pig. In size and in structure they are very similar, particularly if we compare the corresponding regions that are most sensitive to the same frequency (33). For given amplitudes of movement of the footplate of the stapes we should expect rather similar injurious effects on the hair cells and similar probabilities of mechanical failure of the supporting structures. We must remember, however, that the intra-aural reflex was not active in our guinea pigs. Man may enjoy significant protection against tones below 500 cps if his intra-aural reflex is active and does not suffer from too rapid fatigue.

The different relations of AP threshold, CM "threshold" and "maximum CM voltage" to the observed anatomical injuries serves as a caution against any simple assumptions as to tests for susceptibility to acute or chronic acoustic trauma. It is frequently suggested that the ease with which an ear can be "fatigued" can be used for this purpose. But we do not know whether the "fatigue" measured in a psychophysical experiment depends on a reduction in the DC

potential of the scala media, on a reduced efficiency of a mechano-electric modulator, on a detachment of the tectorial membrane from the sensory "hairs," on a change in electrical conductivity of the organ of Corti, on exhaustion of reserve materials in the hair cells, on a rise in the threshold of the endings of the auditory nerve, or a combination of these and other possible factors. As to acute acoustic trauma, there are at least two different mechanical effects which have been caused by our intense brief exposures. First, the microscopic structure within the hair cells is disrupted, and second, one or another of the supporting structures breaks, just as any solid material breaks after a sufficient number of bendings beyond its elastic limit. Furthermore these acute effects may be quite different from whatever cumulative effects lead to deafness after weeks or months of alternate "fatigue" and recovery. These chronic cumulative injuries must be investigated separately and directly. The present study has been concerned only with structural failures at either the microscopic or the sub-microscopic level, that are caused by severe but brief acoustic stresses.

We have postulated an "invisible effect" as the basis of the reduction of CM voltage and elevation of CM threshold in cases in which the hair cells were relatively normal but where there was a near-by rupture of the organ of Corti or of Reissner's membrane. It is well known that such an unexplained depression of electrical output regularly follows any surgical injury to scala media except the insertion of the very smallest microelectrodes. We suggest two possible types of such invisible effects, one chemical the other electrical.

The chemical composition of the endolymph is very different from that of both perilymph and serum in that it has a much higher concentration of potassium and a much lower concentration of sodium (33). Its ionic composition is unique among the body fluids but very much like that of protoplasm. It seems plausible that a major change in the ionic content of endolymph caused by dilution by perilymph entering through a hole in Reissner's membrane or serum leaking from a surgical wound may affect the organ of Corti adversely and depress the production of CM, even though we do not know the exact details involved.

Another explanation of the depression of CM depends on the hypothesis (11,27) that the CM is derived directly from the DC potential of the scala media discovered by Bekesy (35). Bekesy points out (36) that this potential can exist only because of the electrical insulating properties of Reissner's

membrane. His argument applies equally well to the cells of Claudius and of Hensen. In fact a rupture anywhere in the walls of the scala media will produce an electrical leak or short circuit and reduce the DC potential within. This, we believe, would directly and automatically reduce the voltage of the cochlear microphonic also. Of course, these chemical and electrical explanations are not mutually exclusive.

APPENDIX

EXPOSURES AND INJURIES

	185 CPS					TURN I DIFFERENTIAL ELECTRODES			
	EXPOSURE		WORST INJURY			LOCAL INJURY		DB LOSS (CM)	
No.	Db ear-drum	Min.	Turn	Hair	Other	Hair	Other	Threshold 8000 and 2000 cps	Voltage 2000 cps 125 db
390	142	1.0	IV 1[1]	4	C[2]	1		8	7
393	142	5.0	III	2		1+		40	18
386	145	1.0	IV	4	R	2	R	43	18
394	145	0.5						3	4
		+5.0	III	3+	R	1+	R	15	8
395	148	0.17		not fixed				12	2
388	148	0.5	IV, III	4		2		11	10
389	148	0.5		not fixed				15	9
387	148	0.5	III, IV	5	R	3+		>60	large
385	148	1.0	III, IV	4,5	R	2+	R	39	30
399	148	0.2						28	8
		+1.0		not fixed				30	8
		+1.0						32	5
397	151	0.08	probable rupture			not		4	3
		+0.33	of eardrum			fixed		4	5
		+0.33						16	8
		+0.17						16	8
396	151	0.08	III	5	Op[3]			6	5
		+0.5	II	5	C	2+		>60	>30
382	151	1.0		not fixed				>60	no data

[1] l and u refer to lower and upper lower parts of a turn and are used when a severe injury is sharply localized.

[2] C and R indicate rupture of the organ of Corti and of Reissner's membrane respectively. (C) indicates partial rupture.

[3] Op indicates a severe operative injury caused in placing an electrode. (Op) indicates probable injury from the operation.

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EXPOSURES AND INJURIES

	545 CPS EXPOSURE		WORST INJURY			TURN I DIFFERENTIAL ELECTRODES LOCAL INJURY		DB LOSS (CM)	
No.	Db ear-drum	Min.	Turn	Hair	Other	Hair	Other	Threshold 8000 and 2000 cps	Voltage 2000 cps 125 db
366	139	2.0		not fixed				11	8
374	140	5.0		not fixed				7	7
373	141	0.5 +5.0	II 1 to IV	3+		1+		14 13	7 10
368	144	1.0	III u	3	R	1+		4	5
369	144	2.0	III,IV	4	Op	1+		35	29
360	145	0.75	III u	4		1+		12	9*
375	142**	1.5	II,III	2+		1+		10	6
372	142	0.5 +5.0	III 1	4		3		6 >60	6 >40
367	144	1.0	II	3+		1+		10	7
370	144	1.0	I to III	3		2		33	no data
340	145	0.5	III 1	5		1+		18	-2
358	146	0.5	III u, IV 1	3+		1+		37	15
350	146	0.33	II,III	5	C	2+		17	14

*360 - Electrodes were very near round window.

**375 - The intensity varied more than usual during this exposure.

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EXPOSURES AND INJURIES

	2000 CPS EXPOSURE		WORST INJURY			TURN I DIFFERENTIAL ELECTRODES LOCAL INJURY		DB LOSS (CM)	
No.	Db ear-drum	Min.	Turn	Hair	Other	Hair	Other	Threshold 8000 and 2000 cps	Voltage 2000 cps 125 db
235	133	1.33	II	1+		1+		3	no data
238	138	0.33	II 1	5	C (Op)[3]	2+		19	no data
239	138	0.5	II 1	2		1+		5	0
245	138	0.5	I m	3	R in II	3		42	20
240	138	1.0	II 1	2		1+		6	6
241	138	1.5	II, III	2		1		15	6
237	138	2.0	I mu	3+		3+		>60	no data
242	139	1.0	II 1	5	C, R	3		>60	32
251	139[4]	0.33						15	no data
	139[4]	+1.00						18	9
	142[4]	+1.00	I mu, II 1	2+	R (Op)[3]	2	R(Op)	19	6
255	142[4]	1.0	II 1	3+		1+		8	7
268	143	0.08			not	fixed		4	9
270	143	0.25	II 1	3	(C)[2]	1+		3	4

[4] 251, 255 - These values may have been 137 and 140 db respectively. The calibration was uncertain for these two experiments.

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EXPOSURES AND INJURIES

	2000 CPS EXPOSURE		WORST INJURY			TURN I DIFFERENTIAL ELECTRODES LOCAL INJURY		DB LOSS (CM)	
No.	Db ear-drum	Min.	Turn	Hair	Other	Hair	Other	Threshold 8000 and 2000 cps	Voltage 2000 cps 125 db
269	143	0.5	II 1	4		1+		22	13
294	143	1.0			not	fixed		9	4
283	143	1.5			not	sectioned		10	10
285	143	2.0	II 1	2	(C)[2]	1+		18	9
271	143	2.0	IV 1	2	R in I u	1+		11	9
281	143	2.0			not	sectioned		>60	>25
290	143	3.0	II u, III 1	4	R	2+		20	12
276	143	3.0			not	sectioned		18	8
279	143	3.0			not	sectioned		>60	>40
287	143	4.0	II 1	2		1+		10	8
293	143	6.0	III 1	4	R	4	R	49	30
273	143	6.0	II	5		3+		>60	>40
267	146	0.08	II,III	4		2		55	26
305	146	0.08			not	fixed		10	1
265	146	0.17	II 1	5	C	1+		9	14
307	146	0.25			not	fixed		6	6
266	146	0.05 +0.17	III u IV 1	5	C	1+		0 4	0 9

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EXPOSURES AND INJURIES

	2000 CPS					TURN I DIFFERENTIAL ELECTRODES		
	EXPOSURE		WORST INJURY			LOCAL INJURY	DB LOSS (CM)	
No.	Db ear-drum	Min.	Turn	Hair	Other	Hair Other	Threshold 8000 and 2000 cps	Voltage 2000 cps 125 db
262	146	0.5	II	2		1+	10	6
347	146	0.5	II 1	2		1+	0	1
308	146	0.5	II 1	4	R	2	5	3
295	146	1.0	I u, II 1	2		1+	5	5
296	146	1.0	II u	3	R	2	23	15
297	146	1.0	II 1	5	C	2	>60	>40
299	146	1.0			not	fixed	37	21
	8000 CPS							
260	141	1.5			specimen lost		26	9
363	143	1.0	II	2 to 4[5]		2 to 4[5]	51	34
448	146	1.0	I u	2		1+	32	22
351	146	1.0	I m,u	2 to 4[5]		1 to 4[5]	42	28
392	146	1.0 +1.0			not	fixed	33 44	23 35

[5]363 and 351 showed "spotty" injury, i.e. normal cells alternating with seriously injured cells.

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EXPOSURES AND INJURIES

	545 CPS		TURN II DIFFERENTIAL ELECTRODES			
	EXPOSURE		LOCAL INJURY		DECIBEL LOSS (CM)	
No.	Db ear-drum	Min.	Hair	Other	Threshold 8000 and 2000 cps	Voltage 2000 cps 125 db
373	141	0.5 +5.0	3		12 38	13 25
368	144	1.0	2	R at II u	29	5
369	144	2.0	1+	III Op	>60	>30
360	145	0.75	Electrodes in II and III 3+	III u = 4	>60	>30
	2000 CPS					
308	146	0.5	2+	R (Op)	30	18
	185 CPS			TURN III		
					500 and 250 cps	1000 and 500 cps at 125 db
390	142	1.0	3	C in IV	>60	>16
393	142	5.0	2		58	10
386	145	1.0	3		37	7
394	145	0.5 +5.0	3+	R	25 >60	17 >30
395	148	0.17	not	fixed	>60	29
388	148	0.5	3		45	22

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EXPOSURES AND INJURIES

	185 CPS		TURN III DIFFERENTIAL ELECTRODES		
	EXPOSURE		LOCAL INJURY	DECIBEL LOSS (CM)	
No.	Db ear-drum	Min.	Hair Other	Threshold 500 and 250 cps	Voltage 1000 and 500 cps - 125 db
389	148	0.5	not fixed	>36	>16
387	148	0.5	5 C	>60	large
385	148	1.0	3 collapse of rods	47	20
399	148	0.2 +1.0 +1.0	not fixed	43 49 56	5 8 15
397	151	0.08 +0.33 +0.33 +0.33	probable rupture of eardrum	19 8 30 35	8 8 12 14
396	151	0.58	5 Op	no data	
382	151	1.0	not fixed	>60	no data
	545 CPS			500 and 250 cps	500 cps 125 db
366	139	2.0	not fixed	8	10
375	142	1.5	2	35	16
372	142	0.5 +5.0	4	46 >60	distorted
370	144	1.0	2 Op	>60	>30
340	145	0.5	2+ C	>60	>40

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EXPOSURES AND INJURIES

	2000 CPS EXPOSURE		TURN III DIFFERENTIAL ELECTRODES LOCAL INJURY		DECIBEL LOSS (CM)	
No.	Db ear-drum	Min.	Hair	Other	Threshold 500 and 250 cps	Voltage 500 cps 125 db
347	146	0.5	1+		18	9
	545 CPS		**TURN IV**			
					250 and 185 cps	500 cps 125 db
374	140	5.0	no specimen		21	9
367	144	1.0	2	atypical	32	4
358	146	0.5	3		46	26
350	146	0.33	3+	C in III	>60	>40

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EXPOSURES AND INJURIES

	2000 CPS EXPOSURE		TURN II REFERENCE TO NECK LOCAL INJURY		DECIBEL LOSS (CM)	
No.	Db ear-drum	Min.	Hair	Other	Threshold 500 and 250 cps	Voltage 500 cps 125 db
235	133	1.33	2		no data	
238	138	0.33	5	C Op?	46	no data
239	138	0.50	2		22	7
245	138	0.50	2	R Op?	26	6
240	138	1.0	2		37	10
241	138	1.5	2		24	13
237	138	2.0	2	Op?	58	no data
242	139	1.0	5	C Op	59	26
251	139 ↓ 139 ↓ 142 ↓	0.3 +1.0 +1.0	 2+		16 27 35	+ 2 2
255	142 ↓	1.0	3+		37	12
268	143	0.08	no	specimen	4	12
270	143	0.25	3	C beginning	19	6
269	143	0.50	2	Deiter's detached	36	18
294	143	1.0	not	fixed	45	15
283	143	1.5	not	sectioned	>60 for pip 22 for tone	
285	143	2.0	2	Deiter's detached	45	10
271	143	2.0	2		33	8

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	2000 CPS EXPOSURE		TURN II REFERENCE TO NECK LOCAL INJURY		DECIBEL LOSS (CM)	
No.	Db ear-drum	Min.	Hair	Other	Threshold 500 and 250 cps	Voltage 500 cps 125 db
281	143	2.0	not sectioned		>60	>30
290	143	3.0	4	R atypical	36	16
276	143	3.0	not sectioned		1	11
279	143	3.0	"	"	>60	>40
287	143	4.0	2		24	11
293	143	6.0	4	R atypical	>60	>40
273	143	6.0	5	C	>60	>40
267	146	0.08	4		>60	>25
305	146	0.08	no specimen		18	6
265	146	0.17	5	C	>60	>23
307	146	0.25	no specimen		15	9
266	146	0.05 +0.17	2+	C in III u	5 38	6 14
262	146	0.5	2	$(Op)^3$	47	35
347	146	0.5	2		no data	
295	146	1.0	2		20	6
296	146	1.0	2+	R	>60	>20
297	146	1.0	5	C	>60	>40
299	146	1.0	no specimen		>60	>26

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EXPOSURES AND INJURIES

	2000 CPS EXPOSURE		TURN IV REFERENCE TO NECK LOCAL INJURY		DECIBEL LOSS (CM)	
No.	Db ear-drum	Min.	Hair	Other	Threshold 500 cps	Voltage 500 cps 125 db
235	133	1.33	1+		9	no data
238	138	0.33	2		13	" "
239	138	0.5	1+		1	" "
245	138	0.5	1+		15	" "
240	138	1.0	1+		18	" "
241	138	1.5	2		10	" "
237	138	2.0	2		31	" "
242	139	1.0	2		1	" "
251	139	0.33			8	" "
	139	+1.00			12	" "
	142	+1.00	1+		14	" "
255	142	1.00	1+		8	" "
268	143	0.08	no specimen		-6	" "
270	143	0.25	?		5	" "
269	143	0.50	1+	C beginning in II	43	" "
271	143	2.0	2		13	" "
281	143	2.0	not sectioned		1	" "

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	2000 CPS EXPOSURE		TURN IV REFERENCE TO NECK LOCAL INJURY		DECIBEL LOSS (CM)	
No.	Db ear-drum	Min.	Hair	Other	Threshold 500 cps	Voltage 500 cps 125 db
279	143	3.0	not sectioned		12	no data
273	143	6.0	2	C in II	>60	" "
267	146	0.08	2		>60	" "
265	146	0.17	2		>60	>30
266	146	0.02 +0.17	2		3 44	6 >35
262	146	0.5	3		>60	>30

BIBLIOGRAPHY

1. Covell, W. P. and Eldredge, D. H. Injury to animal ears by intense sound. AF Technical Report No. 6561, 1951.

2. Lurie, M. H., Davis, H. and Hawkins, J. E. Acoustic trauma of the organ of Corti in the guinea pig. Laryngoscope, 1944, 54: 375-386.

3. Smith, K. R. The problem of stimulation deafness. II. Histological changes in the cochlea as a function of tonal frequency. J. exp. Psychol., 1947, 37: 304-317.

4. Alexander, I. E. and Githler, F. J. Histological examination of cochlear structure following exposure to jet engine noise. J. comp. and Physiol. Psychol., 1951, 44: 513-524.

5. Davis, H., Gernandt, B. E., Riesco-MacClure, J. S. and Covell, W. P. Aural microphonics in the cochlea of the guinea pig. J. acous. Soc. Amer., 1949, 21: 502-510.

6. Davis, H., Gernandt, B. E. and Riesco-MacClure, J. S. Threshold of action potentials in ear of guinea pig. J. Neurophysiol., 1950, 13: 73-87.

7. Tasaki, I. and Fernández, C. Modification of cochlear microphonics and action potentials by KCl solution and by direct currents. J. Neurophysiol., 1952, 15: 497-512.

8. Tasaki, I., Davis, H. and Legouix, J.-P. The space-time pattern of the cochlear microphonics (guinea pig), as recorded by differential electrodes. J. acous. Soc. Amer., 1952, 24: 502-519.

9. Fernández, C., von Gierke, H., Eldredge, D. H., Jr. and Davis, H. Electrical studies of injury produced in guinea pig's ear by strong high-frequency sounds. Amer. J. Physiol., 1950, 163: 710.

10. Davis, H., Silverman, S. R. and McAuliffe, D. R. Some observations on pitch and frequency. J. acous. Soc. Amer., 1951, 23: 40-42.

11. Davis, H., Tasaki, I. and Goldstein, R. The peripheral origin of activity, with reference to the ear. Cold Spring Harbor Symposia on Quantitative Biology, 1952, in press.

12. Benson, R. W. The calibration and use of probe-tube microphones. J. acous. Soc. Amer., 1953, 25: 128-134.

13. Wiggers, H. The functions of the intra-aural muscles. Amer. J. Physiol., 1937, 120: 781-797.

14. Guild, S. R. War deafness and its prevention -- report of the labyrinths of the animals used in testing of preventive measures (middle ears previously reported). J. Lab. and Clin. Med., 1919, 4: 153-180.

15. Rüedi, L. and Furrer, W. Special kind of acoustic trauma produced by jet engines. A.M.A. Arch. Otolaryngol., 1951, 54: 534-541.

16. Rüedi, L. and Furrer, W. Das akustische Trauma. Karger, Basel, 1947.

17. Hilding, A. C. Studies on the otic labyrinth. I. On the origin and insertion of the tectorial membrane. Ann. Otol., Rhinol. and Laryngol, 1952, 61: 354-370.

18. Katsuki, Y. and Covell, W. P. The organ of Corti by phase-contrast microscopy. The Laryngoscope, 1953, 63: 1-17.

19. Davis, H., Derbyshire, A. J., Kemp, E. H., Lurie, M. H. and Upton, M. Functional and histological changes in the cochlea of the guinea pig resulting from prolonged stimulation. J. Gen. Psychol., 1935, 12: 251-278.

20. Lawrence, M. and Wever, E. G. Effects of oxygen deprivation upon the structure of the organ of Corti. A.M.A. Arch. Otolaryngol., 1952, 55: 31-37.

21. Krejci, F. and Bornschein, H. Der Einfluss chronischer Larmschadigung auf die Cochlearpotentiale von Meerschweinchen. Acta Oto-Laryngol., 1952, 39: 68-79.

22. Tonndorf, J. and Brogan, F. A. Two forms of change in cochlear microphonics; parallel shift in stimulus intensity and truncation of gradient curves; and Cochlear microphonics following noise exposure. School of Aviation Medicine, Project Number 21-27-001, Reports Nos. 6 and 7, 1952.

23. Alexander, I. E. and Githler, F. J. The effects of jet engine noise on the cochlear response of the guinea pig. J. comp. Physiol. Psychol., 1949, 42: 517-525.

24. von Gierke, H. E. Subharmonics generated in ears of humans and animals at intense sound levels. Fed. Proc., 1950, 9: 130.

25. McAuliffe, D. R. Mathematical and electrical analysis of pulsed tones. Thesis, Washington University, 1952.

26. Schuknecht, H. F. and Neff, W. D. Hearing losses after apical lesions in the cochlea. Acta Oto-Laryngol., 1952, 42: 263-274.

27. Davis, H. and Tasaki, I. A possible mechanism of excitation of nerve impulses in the cochlea. Science, 1952, 116: 519-520.

28. Wever, E. G. and Lawrence, M. The patterns of response in the cochlea. J. acous. Soc. Amer., 1949, 21: 127-134.

29. Wing, K. G., Harris, J. D., Stover, A. D. and Brouillette, J. H. Effects of changes in arterial oxygen and carbon dioxide upon cochlear microphonics. Medical Research Laboratory Report No. 188, Bureau of Medicine and Surgery, Navy Department, Project NM 003 041.27.03, 1952.

30. Riesco-MacClure, J. S., Davis, H., Gernandt, B. E. and Covell, W. P. Ante-mortem failure of the aural microphonic in the guinea pig. Proc. Soc. Exp. Biol., 1949, 71: 158-160.

31. Goldstein, R. Reversals of cochlear response patterns with death of the animal. Amer. J. Physiol., 1950, 163: 715. A study of cochlear potentials. Thesis, Washington University, 1952.

32. Davis, H., Morgan, C. T., Hawkins, J. E., Jr., Galambos, R. and Smith, F. W. Temporary deafness following exposure to loud tones and noise. Acta Oto-Laryngol., 1950, Supplement 88.

33. Fernández, C. Dimensions of the cochlea (guinea pig). J. acous. Soc. Amer., 1952, 24: 519-523.

34. Smith, C. A., Wu, M. L. and Lowry, O. H. The electrolytes of the endolymph and perilymph. Science, 1952, 116: 529.

35. Békésy, G. von. DC potentials and energy balance of the cochlear partition. J. acous. Soc. Amer., 1951, 23: 576-582.

36. Békésy, G. von. Gross localization of the place of origin of the cochlear microphonics. J. acous. Soc. Amer., 1952, 24: 399-409.